national Application No T/DK2004/000548

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| A. CLASSI IPC 7 | FICATION OF SUBJECT MATTER C12Q1/34 C12Q1/40 | | _ |
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| | o International Patent Classification (IPC) or to both national classifica | tion and IPC | |
| B. FIELDS | SEARCHED cumentation searched (classification system followed by classification) | un eymbols) | |
| IPC 7 | C12Q | נפוטעוניקט זיי | |
| Documental | tion searched other than minimum documentation to the extent that so | uch documents are included in the fields se | earched |
| Electronic d | ata base consulted during the international search (name of data bas | se and, where practical, search terms used |) |
| EPO-In | ternal, WPI Data, PAJ, CHEM ABS Data | , EMBASE, BIOSIS | |
| C. DOCUM | ENTS CONSIDERED TO BE RELEVANT | | |
| Category ° | Citation of document, with indication, where appropriate, of the rele | evant passages | Relevant to claim No. |
| X | HANSEN S U ET AL: "Direct NMR-spectroscopic determination o active-enzyme concentration by ti with a labeled inhibitor: determi | tration | 1–5 |
| Υ | the k(cat) value of almond beta-glucosidase." CHEMBIOCHEM: A EUROPEAN JOURNAL CHEMICAL BIOLOGY. 2 OCT 2000, vol. 1, no. 3, 2 October 2000 (2000-10-02), page 177-180, XP002310764 ISSN: 1439-4227 page 177, right-hand column, last paragraph - page 178, left-hand c paragraph 3; figure 4; tables 1,2 page 180, left-hand column, last | OF es column, | 6,7 |
| | - | -/ | |
| X Furt | ther documents are listed in the continuation of box C. | X Patent family members are listed | in annex. |
| ° Special co | alegories of cited documents : | *T* later document published after the inte | |
| "A" docum | ent defining the general state of the art which is not dered to be of particular relevance | or priority date and not in conflict with cited to understand the principle or th | the application but |
| | document but published on or after the international | invention "X" document of particular relevance; the | |
| "L' docum | ent which may throw doubts on priority claim(s) or is cited to establish the publication date of another | cannot be considered novel or canno involve an inventive step when the do | cument is taken alone |
| citatio | ns criber to establish the publication date of allottier on or other special reason (as specified) nent referring to an oral disclosure, use, exhibition or | "Y" document of particular relevance; the cannot be considered to involve an indocument is combined with one or m | ventive step when the |
| other | neans of all oral disclosure, use, exhibition or means the means ent published prior to the international filing date but | ments, such combination being obvio in the art. | |
| later t | han the priority date claimed actual completion of the international search | *&" document member of the same patent | |
| 1 | 24 February 2005 | Date of mailing of the international sea | исп героп |
| <u></u> | | | |
| Name and | mailing address of the ISA European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax (+31-70) 340-3016 | Authorized officer Marttin, E | |

:ional Application No

| | | Fe1/DK2004/000548 |
|-----------|--|-----------------------|
| .(Continu | ation) DOCUMENTS CONSIDERED TO BE RELEVANT Citation of document, with indication, where appropriate, of the relevant passages | Relevant to claim No. |
| | The second secon | |
| X | IM H ET AL: "Characterization of high pI alpha-glucosidase from germinated barley seeds: substrate specificity, subsite affinities and active-site residues" CARBOHYDRATE RESEARCH, ELSEVIER SCIENTIFIC PUBLISHING COMPANY. AMSTERDAM, NL, vol. 277, no. 1, 7 November 1995 (1995-11-07), pages 145-159, XP004021839 ISSN: 0008-6215 page 148, last paragraph - page 149, paragraph 3 page 151, paragraph 2 - page 152, paragraph 2; figure 2 page 156, line 4 - line 6 | 1,2,4,5, |
| | STREET I P ET AL: "Inactivation of a beta-glucosidase through the accumulation of a stable 2-deoxy-2-fluoro-alpha-D-glucopyranosyl-en zyme intermediate: a detailed investigation." BIOCHEMISTRY. 20 OCT 1992, vol. 31, no. 41, 20 October 1992 (1992-10-20), pages 9970-9978, XP002319052 ISSN: 0006-2960 page 9970, left-hand column, last paragraph - page 9971, left-hand column, paragraph 2 page 9973, right-hand column, last paragraph - page 9974, left-hand column, paragraph 1; figure 3 page 9977, right-hand column, last paragraph | 1,3-5 |
| Υ | WO 01/32844 A (NOVOZYMES AS) 10 May 2001 (2001-05-10) cited in the application page 37, line 8 - last line page 40, line 30 - page 42, line 5; claims 1,10,14,15; examples 2,4 | 6,7 |
| A | A. CORNISH-BOWDEN: "Fundamentals of enzyme kinetics" 1995, PORTLAND PRESS, LONDON, XP002310765 ISBN: 1 85578 072 0 * revised edition, chapter 11.2, page 274 - page 277 the whole document | 1,2,4-9 |

in tional Application No

| | | TC1/DK2004/000548 |
|-------------|--|-----------------------|
| C.(Continua | ation) DOCUMENTS CONSIDERED TO BE RELEVANT | |
| Category ° | Citation of document, with Indication, where appropriate, of the relevant passages | Relevant to claim No. |
| A | SERI K ET AL: "1-Arabinose selectively inhibits intestinal sucrase in an uncompetitive manner and suppresses glycemic response after sucrose ingestion in animals" METABOLISM, CLINICAL AND EXPERIMENTAL, W.B. SAUNDERS CO., PHILADELPHIA, PA, US, vol. 45, no. 11, November 1996 (1996-11), pages 1368-1374, XP004538505 ISSN: 0026-0495 table 1 | 9 |
| A | EP 0 104 780 A (WAKO PURE CHEM IND LTD) 4 April 1984 (1984-04-04) the whole document | 1,5,8 |
| Α . | WO 02/10439 A (HORTIN GLEN L ;GOVERNMENT OF UNITED STATES OF (US)) 7 February 2002 (2002-02-07) abstract | 1,5,8 |
| A | US 6 162 614 A (EMI SHIGENORI ET AL) 19 December 2000 (2000-12-19) abstract | 1,5,8 |
| A . | EP 0 048 989 A (CORNELL RES FOUNDATION INC) 7 April 1982 (1982-04-07) claims; examples | 1,5,8 |
| A | D ROTTICCI ET AL: "An active-site titration method for lipases" BIOCHIMICA ET BIOPHYSICA ACTA, vol. 1483, 3 January 2000 (2000-01-03), pages 132-140, XP002266893 the whole document | 1,5,8 |
| | | |

rnational application No. PCT/DK2004/000548

| Box II | Observations where certain claims were found unsearchable (Continuation of Item 2 of first sheet) |
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| This Inte | ernational Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons: |
| 1. | Claims Nos.: because they relate to subject matter not required to be searched by this Authority, namely: |
| 2. X | Claims Nos.: because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful international Search can be carried out, specifically: See FURTHER INFORMATION sheet PCT/ISA/210 |
| з | Claims Nos.: because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a). |
| Box II | Observations where unity of invention is lacking (Continuation of item 3 of first sheet) |
| This In | ernational Searching Authority found multiple inventions in this international application, as follows: |
| | see additional sheet |
| 1. X | As all required additional search fees were timely paid by the applicant, this international Search Report covers all searchable claims. |
| 2. | As all searchable claims could be searched without effort Justifying an additional fee, this Authority did not invite payment of any additional fee. |
| з | As only some of the required additional search fees were timely pald by the applicant, this international Search Report covers only those claims for which fees were paid, specifically claims Nos.: |
| 4. | No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the Invention first men tioned in the claims; it is covered by claims Nos.: |
| Rema | The additional search fees were accompanied by the applicant's protest. X No protest accompanied the payment of additional search fees. |

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box II.2

Claims Nos.: -

Present claims 1-7 relate to compounds defined by reference to desirable characteristics or properties, namely "having a Kd which is at least 25 times lower than the concentration of glycosyl hydrolase" and "a substrate wherein the rate constant for the glycosylation step is at least 10 times larger than for the deglycosylation step". The term "having a Kd which is at least 25 times lower than the concentration of glycosyl hydrolase" as used in the present independent claims 1 and 6 and in dependent claim 2 defines an inhibitor by a rate constant. The terms "a substrate wherein the rate constant for the glycosylation step is at least 10 times larger than for the deglycosylation step" as used in the present independent claims 1 and 6 and in dependent claim 3 defines a substrate by a rate constant. However, a compound cannot be sufficiently characterised by a rate constant as it is done by such expressions, because it is impossible to know which substances are encompassed in these expressions.

The claims cover all compounds having this characteristic or property, whereas the application provides support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT for only a very limited number of such compounds. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible. Independent of the above reasoning, the claims also lack clarity (Article 6 PCT). An attempt is made to define the compound by reference to a result to be achieved. Again, this lack of clarity in the present case is such as to render a meaningful search over the whole of the claimed scope impossible. Consequently, the search has been carried out for those parts of the claims which appear to be clear, supported and disclosed, namely those parts relating to the inhibitors mentioned in the description at page 4, line 4 and lines 14-27 and the compounds used in examples 1 and 2, and the concepts of an inhibitor having a Kd which is at least 25 times lower than the concentration of glycosyl hydrolase, and a substrate wherein the rate constant for the glycosylation step is at least 10 times larger than for the deglycosylation step.

The applicant's attention is drawn to the fact that claims relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure. If the application proceeds into the regional phase before the EPO, the applicant is reminded that a search may be carried out during examination before the EPO (see EPO Guideline C-VI, 8.5), should the problems which led to the Article 17(2) declaration be overcome.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 1 in part, 2, 4 in part, 5, 6 in part, 7 in part, 8 and 9

A method for determining the concentration of a glycosyl hydrolase by active-site titration using an inhibitor having a Kd which is at least 25 times lower than the concentration of glycosyl hydrolase; and a method of screening for a property of a glycosyl hydrolase comprising determining the concentration of the glycosyl hydrolase with said method; and the use of acarbose in active-site titration of a glycosyl hydrolase.

2. claims: 1 in part, 3, 4 in part, 6 in part, 7 in part

A method for determining the concentration of a retaining glycosyl hydrolase using a substrate wherein the rate constant for the glycosylation step is at least 10 times larger than for the deglycosylation step; and a method of screening for a property of a glycosyl hydrolase comprising determining the concentration of the glycosyl hydrolase with said method.

information on patent family members

itional Application No

| Patent document cited in search report | | Publication date | | Patent family member(s) | Publication date |
|--|-------|---------------------|----|----------------------------|------------------|
| WO 0132844 | Α | 10-05-2001 | AU | 7646200 A | 14-05-2001 |
| 110 01010 | •• | 10 00 2001 | CA | 2388982 A1 | 10-05-2001 |
| | | | WO | 0132844 A1 | 10-05-2001 |
| | | | EP | 1230348 A1 | 14-08-2002 |
| | | | | | 17 00 2002 |
| EP 0104780 | Α | 04-04-1984 | JР | 1736853 C | 26-02-1993 |
| | • • | | JP | 4024040 B | 23-04-1992 |
| | | | JP | 60030697 A | 16-02-1985 |
| | | | ĴΡ | 1643637 C | 28-02-1992 |
| | | | ĴΡ | 2005400 B | 01-02-1990 |
| | | | ĴΡ | 59039300 A | 03-03-1984 |
| | | | ĀT | 23880 T | 15-12-1986 |
| | | | DE | 3367936 D1 | 15-01-1987 |
| | | | EP | 0104780 A1 | 04-04-1984 |
| | | | | | |
| WO 0210439 | Α | 07-02-2002 | ΑU | 8539201 A | 13-02-2002 |
| | | | WO | 0210439 A2 | 07-02-2002 |
| UC 6160614 | | 10 10 0000 | | 1010400 4 | 05 10 1000 |
| US 6162614 | Α | 19-12-2000 | JP | 1319493 A | 25-12-1989 |
| | | | JP | 1953622 C | 28-07-1995 |
| | | | JP | 6076431 B | 28-09-1994 |
| | | | DE | 68928496 D1 | 29-01-1998 |
| | | | DE | 68928496 T2 | 16-04-1998 |
| | | | EP | 0346912 A2 | 20-12-1989 |
| EP 0048989 | A | 07-04-1982 | AT | 15387 T | 15-09-1985 |
| 00.000 | •• | J, U, 2502 | CA | 1176560 A1 | 23-10-1984 |
| | | | DE | 3172143 D1 | 10-10-1985 |
| | | | ĔΡ | 0048989 A2 | 07-04-1982 |
| | | | ĴΡ | 4001304 B | 10-01-1992 |
| | | | JΡ | 57094660 A | 12-06-1982 |
| | | | ٠. | -, 02 1000 11 | 7F 00 1702 |